EFFECT OF SOUND ON ADRENOCORTICAL AND THYROID FUNCTION AND HIGHER NERVOUS ACTIVITY OF ALBINO RATS

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A combination of the action of physical factors of flight with nervous and emotional stress is bound to produce neuro-endocrine changes. /334

One manifestation of the adaptation syndrome is a change in function of the pituitary—adrenal system. Disturbance of its activity through exhaustion following prolonged application of stressor stimuli may lower the body's resistance to the action of unfavorable factors, notably to the factors of space flight. In many published reports data are given for changes in pituitary, adrenal and thyroid functions under the influence of various stressors. However, we know of no investigations to compare the functional state of the pituitary—adrenal system and thyroid gland during stress with the activity of the nervous system. /335

In this paper we describe the results of an experimental study of the effect of sound on adrenocortical and thyroid function and on the higher nervous activity of albino rats. An acoustic stimulus was chosen as stressor for two reasons. First, sound is readily applied in accurately measured intensity, frequency, and duration of action. Second, sound is a physical factor inevitably encountered in actual space flight.

Our experiments were carried out in the autumn and winter on Wistar albino rats weighting 150-210 g. An acoustic stimulus from a type ZG-10 audiofrequency generator, with a frequency of 650 Hz, intensity 70 dB, and duration 17 sec, was applied every 13 sec. The duration of acoustic stimulation varied from 1 hr to 14 days.

RESULTS AND DISCUSSION

Effect of Sound on Adrenocortical Function. The data in the literature relating to the effect of sound on the pituitary—adrenal system are contradictory, evidently because of differences in the technical conditions used in the experiments (different sources of sound, the frequency characteristics, intensity, and duration of action of the stimulus were not always determined, and so on). For instance, Anthony and Babcock [11], who used sound with an intensity of 110-140 dB and with frequencies of between 150-4800 Hz and between 10,000 and 20,000 Hz for 5 min, as a single exposure or daily for 4 weeks, found no changes in the cholesterol content in the adrenals or in their weight, and no changes in concentration of the plasma electrolytes. Duncan [14] observed changes in the content of corticosteroids and the ascorbic acid concentration in the adrenals after exposure to a bell for 1 min.

Changes in adrenocortical activity indicating activation of the pituitary—adrenal system under the influence of acoustic stimulus have been found by several other investigators [13, 16, 17].

In our experiments we judged adrenocortical function by the content of cholesterol and ascorbic acid in the adrenal cortex, changes in the weight of the gland, and synthesis of corticosteroids during incubation of the gland in vitro. /336
Ascorbic acid was determined by the method of Roe and Kuethner as modified by A. D. Ryzhova [8], cholesterol by the method of M.I. Prokhorova and Z.N. Tupikova [7], and synthesis of corticosteroids by adrenal slices by the method of Van der Vies [18], with certain modifications.

The results of experiments to determine the state of adrenal function are summarized in Table 1. They show that exposure to the sound used in these experiments for 1 h had no immediate effect on adrenocortical function. After exposure to the stimulus for 3 h, the ascorbic acid concentration in the glands fell. Exposure to acoustic stimulation for 6 h reduced the concentration of ascorbic acid and cholesterol. After exposure to sound for 12 h, 24 h, and 3 days, only the cholesterol concentration was reduced. Stimulation for 12 h was accompanied by hypertrophy of the adrenals also. The adrenal function of animals exposed to acoustic stimulation for 8 days was indistinguishable from normal. The absence /337
of changes in adrenal activity in response to acoustic stimulation in this case cannot be explained by exhaustion of the glands, because injection of adrenocorticotropic hormone 3 h before the end of the 8-hour experiment lowered the ascorbic and cholesterol concentrations in the adrenals just as in intact animals.

The dynamics of changes in ascorbic acid and cholesterol concentrations in the adrenals during acoustic stimulation is shown in Fig. 1.

Production of corticosteroids during incubation of adrenal slices under these experimental conditions was unchanged. It is interesting to note that in experiments performed in the summer, but not included in this paper, an increase in synthesis of corticosteroids in vitro was observed after exposure to sound for 6 and 12 h.

Hence, like other investigators, we observed changes in adrenal functions under the influence of sound. Under the experimental conditions used, application of the acoustic stimulus for 3 h activated the adrenals, the effect reaching its maximum after 6-12 h. Absence of appreciable changes in adrenal activity after application of acoustic stimulation for 8 days may evidently be attributed to adaptation of the animal to the action of sound.

Effect of Sound on Thyroid Function. In an investigation by Yu. B. Skebel'skaya [9], the action of acoustic stimulation on thyroid function was studied (a loud electric bell was used, but no indication of the absolute intensity of the sound is given). This investigator states that a single exposure to the bell for 2 min causes changes in the histological picture of the gland capable of interpretation as stimulation of its activity. Application of this stimulus daily for 9 days caused marked inhibition of thyroid function.

During the action of combined stimuli (acoustic and photic) for 5 min every /338
hour and during a single daily exposure to acoustic, photic, and electrodermal

TABLE 1. Effect of Acoustic Stimulation on Adrenocortical Function of Albino Rats

Duration of Acoustic Stimulation	Group	Weight of Adrenals, mg/100 g Body Weight	Production of Corticosteroids by Slices, μg/100 mg/h	Ascorbic Acid, mg% of Moist Weight	Cholesterol, Percent of Moist Weight
1 h	C	21.9∓1.74 (9)	7.6∓1.10 (5)	386∓18 (4)	3.7∓0.19 (4)
	E	22.5∓1.70 (10)	8.6∓1.29 (5)	384∓27 (5)	4.3∓0.27 (5)
3 h	C	19.0∓0.79 (10)	9.7∓0.60 (5)	440∓23 (5)	4.1∓0.13 (5)
	E	16.6∓0.84 (10)	9.5∓0.57 (5)	390∓16 (5)*	4.5∓0.38 (5)
6 h	C	14.8∓0.61 (9)	7.8∓0.44 (4)	413∓11 (9)	4.4∓0.35 (10)
	E	16.2∓0.66 (10)	8.9∓1.13 (5)	319∓14 (10)**	3.2∓0.33 (10)*
12 h	C	21.0∓2.39 (10)	10.3∓1.36 (5)	412∓13 (17)	3.6∓0.26 (18)
	E	30.5∓2.08 (10)**	9.6∓1.81 (4)	335∓16 (18)**	2.3∓0.19 (18)**
1 day	C	24.2∓0.97 (15)	10.1∓0.70 (10)	426∓ 9 (17)	3.7∓0.40 (10)
	E	25.0∓1.92 (14)	8.8∓1.25 (10)	383∓21 (17)*	2.7∓0.59 (10)**
3 days	C	24.2∓1.49 (10)	12.0∓0.81 (10)	495∓18 (10)	3.7∓0.17 (10)
	E	25.7∓1.44 (10)	10.0∓0.96 (10)	475∓21 (10)	2.9∓0.12 (10)**
8 days	C	26.0∓0.45 (20)	11.4∓0.47 (10)	438∓13 (10)	3.5∓0.20 (10)
	E	27.0∓0.73 (20)	11.9∓1.06 (10)	432∓15 (10)	3.5∓0.25 (10)

* $P < 0.05$. ** $P < 0.001$.

Note: Number of Experiments in Parentheses; C Denotes Control, E Denotes Experimental Animals.

stimulation for 5 min, thyroid function was found to be increased. Later it returned to its initial level, and on the following days the activity of the gland was inhibited [10].

Daily exposure for 15 days to the alternate action of a loud noise (bell, buzzer) and to a light (150-W lamp) inhibited thyroid function [4].

Two indices were used to assess thyroid function in our investigations: the level of protein-bound iodine in the blood plasma (PBI) and the histological picture of the thyroid gland (height of the follicular epithelium and character of the colloid).

The PBI level was determined by the method of Barker and co-workers [12] as modified by T.F. Komarova, D.S. Tendler, and Ye.V. Sokolova [3]. In the histological investigation the thyroid was fixed in Bouin's fluid and embedded in paraffin wax. Sections were cut to a thickness of 5-6 μ and stained by Heidenhain's

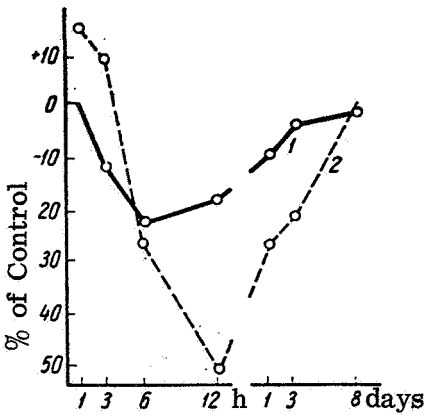

Figure 1. Dynamics of Changes in Ascorbic Acid (1) and Cholesterol (2) Concentrations in Adrenals of Rats During Action of Acoustic Stimulation.

method. To determine the mean height of the thyroid epithelial cells, the height of the cells was measured in sections passing through the center of the gland by means of an ocular micrometer with an immersion objective 90x. Altogether 500 cells were measured in each gland. The results of the measurements were analyzed by statistical methods.

For a period of 4-7 days before the series of experiments began the rats were placed in a room in which the temperature was maintained at 18-20°. Acoustic stimulation lasted for 6-12-18 h and 1, 3, 7, and 14 days. From 5 to 10 experimental and control animals were used for each period of exposure. The plasma PBI level in the rats exposed to sound for 12 and 18 h was the same as in the control animals. In the histological picture of the thyroid gland only a slight increase in height of the follicular epithelial cells with widening of the zones of colloid resorption and the appearance of interfollicular droplets were observed.

Application of the acoustic stimulus for 24 h caused a more distinct and varied effect. As may be seen in Table 2, in four of five experiments a significant increase in the PBI level was observed. In the first two experiments the height of the follicular epithelium was lowered. Demarcation into peripheral and central parts in relation to size of the follicles and of the epithelial cells lining them was sharper in these sections than under normal conditions. The follicles in the parenchyma were mainly regular and oval in shape, and were filled with a thick, sometimes stratified colloid. The follicular epithelial cells were low-cubical or flattened in shape. Their mean height was $7.97 \pm 0.048\,\mu$. Very few /339
undifferentiated interfollicular islets were present, and there were no signs of proliferation of the parenchyma. In the other three experiments no histological changes were observed in the thyroid gland.

Acoustic stimulation for 3, 7, and 14 days had no demonstrable effect on thyroid function.

The fact was noted that changes in the indices used to reflect thyroid activity give conflicting information concerning its state. Whereas the raised plasma PBI level indicates activation of the gland, the reduced height of the follicular epithelium is evidence of inhibition of its function. It will be noted that the use of different criteria for assessing thyroid function, judging by data in the literature, does not always give corresponding results [10, 15]. This is particularly true when the morphological picture of the thyroid is compared with other indices of its functional state.

However, despite the lack of uniformity in the results of our experiments, it can evidently be concluded that prolonged exposure to acoustic stimulation of a

TABLE 2. Changes in Thyroid Function Following Acoustic Stimulation for 24 h

PBI, μg%			Height of Follicular Epithelium, μ		
Control	Experiment, and	P	Control	Experiment, and	P
1,61∓0,113	2,9∓0,072	0,01	9,94∓0,049	8,12∓0,099	0,01
1,79∓0,203	2,21∓0,097	0,1	12,50∓0,063	7,97∓0,048	0,01
2,50∓0,135	1,86∓0,211	0,05	10,66∓0,519	10,27∓0,10	0,6
0,97∓0,068	1,27∓0,146	0,1	13,43∓0,16	13,28∓0,146	0,5
1,86∓0,190	2,69∓0,360	0,1	10,72∓0,470	10,28∓0,071	0,7

Commas represent decimal points.

certain intensity modifies thyroid function. Exposure to sound for 6, 12, and 18 h clearly proved to be a comparatively mild stimulus, causing no distinct changes in thyroid activity. Exposure to sound for 24 h caused an increase in the plasma PBI, indicating increased thyroid function. The decrease in height of the follicular epithelium in these experiments may be attributed to increased secretion of hormone into the blood stream, evidence of which is given by the elevation of the PBI level. The absence of marked changes in thyroid function as a result of the action of the acoustic stimulus for 3, 7, and 14 days may be explained by adaptation of the animal to this stimulus.

An increase in the intensity of the stimulus could possibly give rise to a third phase—a phase of inhibition of thyroid activity. /340

Effect of Sound on Higher Nervous Activity. Experiments were carried out on a separate group of rats consisting of 9 animals, using the motor electrodefensive technique of A.B. Glebovskiy and V.K. Fedorov [2]. A bell was used as conditioned stimulus. The scheme of the experiment was as follows. In the first experiment the animals were first taught to turn to the right in a maze. After they had done this five times in succession without a mistake, the direction of turning was modified. The experiment ended as soon as the rats turned to the left five times in succession. The next experiment began with the rats turning to the left and ended with them turning to the right. This continued throughout the period of investigation. The state of higher nervous activity was judged from the latent period of the motor response, i.e., the time from the beginning of action of the bell until the rat left its circle, the time taken to run through the maze, and the number of mistakes made by the animal during initial learning and modification of the procedure.

The effect of sound was studied three weeks after the beginning of work with the animals, when the indices to be investigated had become constant.

To judge the changes produced by acoustic stimulation, we compared the latent periods, the times taken to run through the maze, and the number of mistakes at three time intervals: 7 experiments before the beginning of acoustic

stimulation, 7 experiments during acoustic stimulation, and 6 experiments after its end. Every day the mean values of these indices were calculated for all 9 rats. The results obtained are given in Table 3.

TABLE 3. Effect of Acoustic Stimulation on Some Indices of Conditioned-Reflex Activity of Rats

Indices Investigated	Period of Investigation			Background-Acoustic Stimulation	Acoustic Stimulation-After Stimulation
	Background	Acoustic Stimulation	After Stimulation		
Latent Period of Training.	7,7±0,85	10,6±0,51	6,4±0,49	$0,02 < P < 0,05$	$P < 0,01$
Time Taken to Run Through Maze.	7,7±1,06	10,7±0,86	8,2±0,64	$0,1 > P > 0,05$	$0,1 > P > 0,05$

Commas represent decimal points.

As the results show, under the influence of sound the latent period of reproduction of the movement was lengthened, and the time taken to run through the maze showed a tendency to increase. The temporal characteristics were modified under the influence of sound in practically all the animals, but a very slight increase in the number of mistakes during reproduction of the movement was observed in only some rats. We consider that the lengthening of the temporal indices is evidence of a state of inhibition in the animals arising under the influence of acoustic stimulation. /341

In some animals the number of mistakes was increased during the first day after discontinuing the sound, possibly indicating adaptation of the animals to this stimulus.

The very slight changes in higher nervous activity which we observed under the influence of acoustic stimulation are not in agreement with the conclusions reached by other workers [1, 5, 6], who found a decrease in magnitude of conditioned reflexes, or their actual disappearance sometimes for long periods, after acoustic stimulation of short duration. The main causes of the discrepancy, in our opinion, are: first, differences in the duration and intensity of the stimuli, and second, the fact that the authors cited used a bell as the acoustic stimulus. This stimulus, which acts specifically on the auditory analyzer of rats, causes them to develop convulsions. In Pavlova's experiments, marked changes in higher nervous activity were observed in rats giving a motor response to ringing of a bell. Monayenkov [5] also describes the variation in changes in higher nervous activity observed among animals "sensitive" and "insensitive" to the ringing of a bell. Wistar rats do not possess specific sensitivity to ringing. To a considerable extent the discrepancy between our findings and those described by the authors cited above may be attributed to different techniques used for studying higher nervous activity. All these authors used Kotlyarevskiy's motor-food method.

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